

# Isoflavone Characterization and Antioxidant Activity of Ohio Soybeans

JaeHwan Lee\*, Marjory Renita#, Steve St. Martin##, Steven. J. Schwartz# and Yael Vodovotz#

## ABSTRACT

Seventeen Ohio soybean cultivars were characterized for their isoflavone and antioxidant activities for possible use as health-promoting ingredients in food products. Isoflavone content was determined by C<sub>18</sub> reverse-phase high performance liquid chromatography (HPLC) coupled with a photodiode array detector. Antioxidant activities of soybean extracts were measured using 2,2-diphenyl-1-picryl-hydrazil (DPPH) and a photochemiluminescence (PCL) methods. Total phenolic compounds (TPCs) in soybean extract were also measured. The highest and lowest total isoflavone content in aglycone equivalent were 304.7 and 111.0 mg/100g soy, respectively, while the average content was 186.3 mg/100g soy. Antioxidant activities of soybean extracts ranged from 7.51 to 12.18 μmol butylated hydroxytoluene (BHT) equivalent/g soy using the DPPH method. Lipid- and water-soluble antioxidant activities of soybean extracts determined using the PCL method ranged from 2.40 to 4.44 μmol Trolox equivalent/g soy and from 174.24 to 430.86 μmol ascorbic acid equivalent/g soy, respectively. No significant correlation between isoflavone content and antioxidant activities were found. TPCs in soybean cultivars showed a weak correlation with total isoflavone content (R<sup>2</sup>=0.40). Five Ohio soybean cultivars containing high isoflavone content and/or high antioxidant activities were identified as possible ingredients for value-added soy-based foods.

## INTRODUCTION

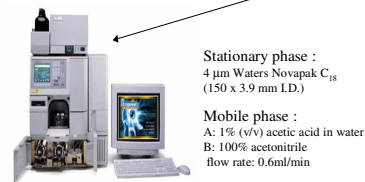
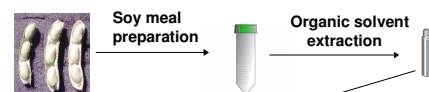
- Consumptions of soybeans and soy products have been associated with reducing the risks of various cancers<sup>1,2</sup>.
- The health promoting activity associated with soy consumption is attributed to the presence of isoflavones. The structural similarities of isoflavones to naturally occurring estrogens may protect hormone-dependent cancers by modulating activity of estrogen<sup>1</sup>.
- Antioxidant properties, especially radical scavenging activities, are important due to the deleterious role of free radicals in foods and in the body.
- Free radicals have been associated with the aging process and age-related diseases, oxidation of lipids in foods, deterioration of food quality and consumer acceptance<sup>3</sup>.
- Western style diet is lacking in acceptable products containing soy<sup>4</sup>.
- Strategy → **increase the use of soy by incorporating soy-based ingredients into traditional products in the Western diet.**
- Selecting for soybean cultivars containing high isoflavone content and/or other health promoting compounds such as antioxidants, may enhance the health benefit effects of food products.
- Soybean is one of the most important crops in Ohio and the leading grain crop in area planted<sup>5</sup>. However, systematic characterizations of isoflavone content and antioxidant activities in Ohio soybean cultivars have not been reported.

## OBJECTIVES

The objectives of this study were to determine the isoflavone content, and antioxidant activities of 17 Ohio soybean cultivars and to identify the soybean cultivars with high isoflavone content and/or high antioxidant activities for further processing into food ingredients of soy-based functional foods.

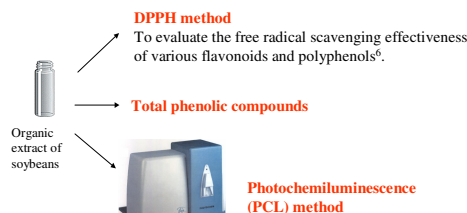
## MATERIALS AND METHOD

### Sample preparation and Isoflavone analysis



### Reversed-phase high performance liquid chromatography with a photodiode array detector (HPLC-PDA)

### Antioxidant measuring methods



'ACW' and 'ACL' are measuring modes for water-soluble and lipid-soluble antioxidant activities, respectively.

PCL method measures the luminescence from luminol, a photosensitizer, that generates superoxide anion when exposed to UV light. Antiradical substances react with the superoxide anion and remaining luminescence is detected<sup>7</sup>.

## RESULTS AND DISCUSSION

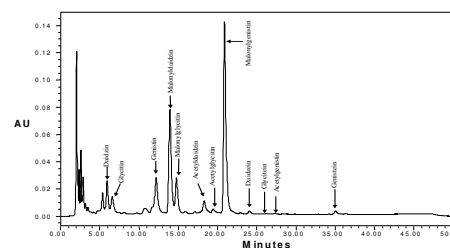


Figure 1. Representative HPLC chromatogram of isoflavones in soybeans. Coefficient of variations for the isoflavone analysis of each soybean variety was less than 5% (n=4).

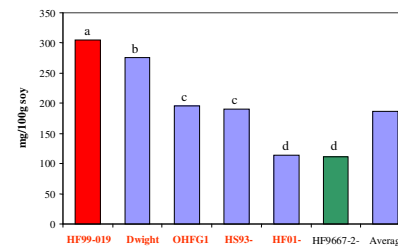


Figure 2. Total isoflavone content of selected soybean varieties.

**The highest and lowest total isoflavone content were 304 and 111 mg/100 g soy, respectively, while the average was 186 mg/100 g soy.**

Table 1. Correlation coefficient (r) among isoflavone content, antioxidant activities, total phenolic compounds in soybeans.

	Total isoflavone Content	DPPH scavenging	a PCL method		Total phenolic compounds
			Lipid-soluble	Water-soluble	
Total isoflavone	*	+0.004	+0.002	+0.049	+0.400
DPPH scavenging		*	+0.027	+0.001	+0.027
Lipid-soluble antioxidant activity			*	+0.394	+0.250
Water-soluble antioxidant activity				*	+0.001

**Data in Table 1 indicated that isoflavones may not be effective free radical or superoxide anion scavengers.**

Table 2. Antioxidant activities and total phenolic compounds of 17 Ohio soybean cultivars.

Cultivars	Antioxidant activities		Total phenolic compounds <sup>‡</sup>
	DPPH method <sup>†</sup>	PCL method	
	Lipidsoluble <sup>‡</sup> water-soluble <sup>‡</sup>		
Dilworth	7.51 <sup>a</sup>	3.19	211.02
Dwight	12.18 <sup>b</sup>	4.44	260.96
HC95-1503	10.82 <sup>b</sup>	4.17	353.46
HF01-0019	9.25 <sup>d</sup>	<b>4.58</b>	<b>430.86</b>
HF02-0218	9.61 <sup>de</sup>	3.55	223.08
HF9662-2-15	9.54 <sup>de</sup>	3.43	203.28
HF9667-2-4	10.14 <sup>de</sup>	3.43	190.00
HF99-019	9.45 <sup>de</sup>	3.42	232.38
HS93-4118	11.79 <sup>b</sup>	3.10	183.96
HS96-3145	10.05 <sup>de</sup>	2.85	320.24
HS96-3850	10.46 <sup>de</sup>	3.07	300.08
HS98-3754	10.59 <sup>de</sup>	3.07	274.44
HS98-3755	10.77 <sup>de</sup>	3.84	338.12
HSO3274	9.73 <sup>de</sup>	2.40	192.24
OHFG1	9.58 <sup>d</sup>	3.76	320.40
OHFG3	10.31 <sup>d</sup>	3.02	191.28
PANA	10.42 <sup>d</sup>	3.00	174.24
Average	10.13	3.43	255.65

\*: unit: μmol BHT equivalent/g soy; †: unit: μmol Trolox equivalent/g soy; ‡: unit: μmol Ascorbic acid equivalent/g soy; §: unit: mg (Catechin equivalent/g soy). Bold character: the highest content among varieties; Italic character: the lowest content among varieties. Coefficient of variation of antioxidant analysis was less than 5%. Different superscripts are significant among varieties (P<0.05).

## CONCLUSION

1. Five Ohio soybean varieties, 'HF99-019', 'Dwight', 'HS93-4118', 'OHFG1' and 'HF01-0019' were identified as possible ingredients for health promoting food products due to their high isoflavone content and/or high antioxidant activities.
2. Isoflavones in soybean may not be an effective form of antioxidants or may not be effective free radical or superoxide anion scavengers.
3. Other compounds soluble in the soybean organic extract may play a role in free radical or superoxide anion scavenging antioxidant activities.

## REFERENCES

1. Hendrich S, G.J. Wang, H.K. Liu, X. Xu, B.Y. Tew, H.I. Wang, and P.A. Murphy. 1999. Isoflavone metabolism and bioavailability, p. 211-230. In A. M. Pappas, ed. Antioxidant Status, Diet, Nutrition, and Health. CRC, Boca Raton, Fla.
2. Kuffing, S.E., D.M. Hong, T.J. Simat, and M. Metzler. 2000. Oxidative in vitro metabolism of soy phytoestrogens daidzein and genistein. J. Agric. Food Chem 48:4963-4972.
3. Wickens, A.P. 2001. Aging and the free radical theory. Respiration Physiology 128:379-391.
4. Klein, B.P., A.K. Perry, and N. Adair. 1995. Incorporation soy proteins into baked products for use in clinical studies. J. Nutrition 125:668-678.
5. USDA. 2002. Oil Crops Situation and Outlook Yearbook. USDA, Washington D.C.
6. Costello, N., J.L. Bernier, J.P. Cateau, J. Poirier, J.C. Walle, and E.M. Geydou. 1996. Antioxidant properties of hydroxy-flavones. Free Radical Biology and Medicine 20:35-43.
7. Popov, L.N., and G. Lewin. 1994. Photochemiluminescent detection of antiradical activity. II. Testing of nonenzymic water-soluble antioxidants. Free Radical Biology and Medicine 17:267-271.

\*Department of Food Science and Technology, Seoul National University of Technology, Seoul, Korea;

#Department of Food Science and Technology, ##Department of Horticulture and Crop Science, The Ohio State University, Columbus, OH 43210, U.S.A.